

**WHAT IS CLAIMED IS:**

1. A method for assessing protein folding and/or solubility comprising:

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- a) providing an expression construct comprising (i) a gene encoding fusion protein, said fusion protein comprising a protein of interest fused to a first segment of a marker protein, wherein said first segment does not affect the folding or solubility of the protein of interest, and (ii) a promoter active in said host cell and operably linked to said gene;
- b) expressing said fusion protein in a host cell that also expresses a second segment of said marker protein, wherein said second segment is capable of structural complementation with said first segment; and
- c) determining structural complementation,

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wherein a greater degree of structural complementation, as compared to structural complementation observed with appropriate negative controls, indicates proper folding and/or solubility of said protein.

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2. The method of claim 1, wherein said fusion is C-terminal to said protein of interest.
3. The method of claim 1, wherein said fusion is N-terminal to said protein of interest.
4. The method of claim 1, wherein said marker protein is selected from the group consisting of a target binding protein, an enzyme, a protein inhibitor, a fluorophore and a chromophore.
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5. The method of claim 4, wherein said marker protein is a target binding protein.
6. The method of claim 5, wherein said target binding protein is ubiquitin.
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7. The method of claim 4, wherein said marker protein is a chromophore.

8. The method of claim 7, wherein said chromophore is green fluorescent protein, blue fluorescent protein, yellow fluorescent protein, luciferase or aquorin.

5 9. The method of claim 4, wherein said marker protein is an enzyme.

10 10. The method of claim 9, wherein said enzyme is  $\beta$ -galactosidase, cytochrome c, chymotrypsin inhibitor, Rnase, phosphoglycerate kinase, invertase, staphylococcal nuclease, thioredoxin C, lactose permease, amino acyl tRNA synthase, and dihydrofolate reductase.

11. The method of claim 10, wherein said enzyme is  $\beta$ -galactosidase.

12. The method of claim 11, wherein said first segment is the  $\alpha$ -peptide of  $\beta$ -galactosidase, and said second segment is the  $\omega$ -peptide of  $\beta$ -galactosidase.

13. The method of claim 1, wherein said protein of interest is Alzheimer's amyloid peptide (A $\beta$ ), SOD1, presenillin 1 and 2,  $\alpha$ -synuclein, amyloid A, amyloid P, CFTR, transthyretin, amylin, lysozyme, gelsolin, p53, rhodopsin, insulin, insulin receptor, fibrillin,  $\alpha$ -ketoacid dehydrogenase, collagen, keratin, PRNP, immunoglobulin light chain, atrial natriuretic peptide, seminal vesicle exocrine protein,  $\beta$ 2-microglobulin, PrP, precalcitonin, ataxin 1, ataxin 2, ataxin 3, ataxin 6, ataxin 7, huntingtin, androgen receptor, CREB-binding protein, dentatorubral pallidoluysian atrophy-associated protein, maltose-binding protein, ABC transporter, glutathione S transferase, and thioredoxin.

14. The method of claim 1, wherein a gene encoding said second segment is carried on a chromosome of said host cell.

15. The method of claim 1, wherein a gene encoding said second segment is carried episomally in said host cell.

16. The method of claim 1, wherein said host cell is selected from the group consisting of a bacterial cell, an insect cell, a yeast cell, a nematode cell, and a mammalian cell.

17. The method of claim 16, wherein said host cell is a bacterial cell.

18. The method of claim 17, wherein said bacterial cell is *E. coli*.

19. The method of claim 18, wherein said promoter is the *Taq* promoter; T7 promoter, or the *P<sub>lac</sub>* promoter.

20. The method of claim 16, wherein said host cell is a nematode cell.

21. The method of claim 20, wherein said nematode cell is a *C. elegans* cell.

22. The method of claim 16, wherein said host cell is an insect cell.

23. The method of claim 22, wherein said host cell is a *S. fugearia* cell.

24. The method of claim 16, wherein said host cell is a yeast cell.

25. The method of claim 14, wherein said promoter is CupADH or Gal.

26. The method of claim 16, wherein said host cell is a mammalian cell.

27. The method of claim 26, wherein said promoter is PepCk or tk.

28. The method of claim 1, wherein said negative control utilizes a host cell lacking the second segment of said marker protein.

29. The method of claim 1, wherein said negative control utilizes a fusion protein that is improperly folded and/or insoluble.

30. A method for screening protein folding and/or solubility mutants comprising:

- a) providing a gene encoding fusion protein comprising (i) a protein of interest and (ii) a first segment of a marker protein, wherein said first segment does not affect the folding or solubility of the protein of interest, , wherein said fusion protein is not properly folded and/or soluble when expressed in said host cell;
- b) mutagenizing that portion of the gene encoding said protein of interest;
- c) expressing said fusion protein in a host cell that expresses a second segment of said marker protein, wherein said second segment is capable of structural complementation with said first segment; and
- d) determining structural complementation,

wherein a relative increase in structural complementation, as compared to the structural complementation observed with the unmutagenized fusion protein, indicates an increase in proper folding and/or solubility of said protein.

31. The method of claim 30, wherein said fusion is C-terminal to said protein of interest.

32. The method of claim 30, wherein said fusion is N-terminal to said protein of interest.

33. The method of claim 30, wherein said marker protein is selected from the group consisting of a target binding protein, an enzyme, a protein inhibitor, a chromophore.

34. The method of claim 30, wherein said host cell is selected from the group consisting of a bacterial cell, an insect cell, a yeast cell, a nematode cell, a mammalian cell.

35. A method for screening candidate modulator substance that modulates protein folding and/or solubility comprising:

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- a) providing an expression construct comprising (i) a gene encoding fusion protein, said fusion protein comprising a protein of interest fused to a first segment of a marker protein, wherein said first segment does not affect the folding or solubility of the protein of interest, and (ii) a promoter active in said host cell and operably linked to said gene;
  - b) expressing said fusion protein in a host cell that expresses a second segment of said marker protein, wherein said second segment is capable of structural complementation with said first segment;
  - c) contacting the host cell with said candidate modulator substance; and
  - 10 d) determining structural complementation,

wherein a relative change in structural complementation, as compared to the structural complementation observed in the absence of said candidate modulator substance, indicates that said candidate modulator substance is a modulator of protein folding and/or solubility.

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- 36. The method of claim 35, wherein said fusion is C-terminal to said protein of interest.
  - 37. The method of claim 35, wherein said fusion is N-terminal to said protein of interest.
  - 38. The method of claim 35, wherein said marker protein is selected from the group consisting of a target binding protein, an enzyme, a protein inhibitor, a chromophore.
  - 39. The method of claim 35, wherein said host cell is selected from the group consisting of a  
25 bacterial cell, an insect cell, a yeast cell, a nematode cell, a mammalian cell.
  - 40. The method of claim 35, wherein said candidate modulator substance is selected from the group consisting of a protein, a nucleic acid or a small molecule.